Development of Vapor Phase Hydrogen Peroxide Sterilization Process for Spacecraft Applications

Naresh Rohatgi, Wayne Schubert, Jennifer Knight, Megan Quigley, Gustaf Forsberg, Gani Ganapathi, Charles Yarbrough, and Robert Koukol
Jet Propulsion Laboratory, California Institute of Technology, California, Pasadena

ABSTRACT

In order to meet microbial reduction requirements for all Mars in-situ life detection and sample return missions, entire planetary spacecraft (including planetary entry probes and planetary landing capsules) may have to be exposed to a qualified sterilization process. At JPL, we are developing a low temperature (~45°C) vapor phase hydrogen peroxide sterilization process. This process is currently being used by the medical industry and its effectiveness is well established.

In order to effectively and safely apply this technology to sterilize a spacecraft, which is made out of various man-made materials and electronic circuit boards, the following technical issues need to be resolved:

1. Efficacy of sterilization process.
2. Diffusion of H_2O_2 under sterilization process conditions into hard to reach places.
3. Materials and components compatibility with the sterilization process.
4. Development of methodology to protect (isolate) sensitive components (i.e. electronic) from H_2O_2 vapor.

This paper will present test data and discussion on the work we are conducting at JPL to address these issues.

I. INTRODUCTION

For interplanetary missions landing on a planet of potential biological interest, planetary protection requires that the flight system must be assembled, tested and ultimately launched with the intent of minimizing the bio-load taken to and deposited on the planet[1,2]. To demonstrate compliance with these requirements, there are specific requirements as to the maximum bio-load of spore forming organisms allowable upon launch from Earth. There are even more restrictive requirements for lander missions, which have a goal of either in-situ life detection or an extraterrestrial sample return to Earth. These requirements strive to ensure that no Earth based organisms are carried along and then find their way into the life detection instrument or the sample to be returned. Historically, such compliance was achieved by sterilizing the flight hardware. In the history of the United States Space program only dry heat (at nominally125°C) has been approved as a sterilization technique[3]. This has been and continues to be a valuable and practical technique for many types of hardware. However, recent performance advances in electronics and other thermally sensitive parts makes using high temperatures unsuitable for some hardware. Another technique utilizing lower temperatures is needed to augment dry heat and provide a viable alternative option. Some of the techniques such as ethylene oxide gas, ultraviolet radiation, paraformaldehyde, and chlorine dioxide were considered[4,5]. These techniques have technical problems such as high corrosivity to spacecraft materials, issues such as toxicity and carcinogenicity, or leaving organic residue on the spacecraft surfaces. The alternative technique selected is vapor phase hydrogen peroxide process. Hydrogen peroxide has been very successfully used in the medical industry with no discernable impact on either the component materials or device performance[6]. The condensation of water and hydrogen peroxide can be easily avoided by maintaining proper process conditions (temperature, pressure and H_2O_2 concentration).

II. VAPOR PHASE H_2O_2 STERILIZATION PROCESS

Sterilizer

The H_2O_2 sterilizer used in this study is the Sterrad® 100 SI GMP Sterilizer manufactured by Advanced Sterilization Products which is a subsidiary of the pharmaceutical company Johnson and Johnson. The sterilizer is shown in Figure 1.

Not shown in Figure 1 are two additional pieces of hardware – the process monitor computer which is a laptop displaying the different process conditions in real

time and a monitor which is hooked up to a built-in computer within the sterilizer. The built-in computer is used to configure the sterilizer for different operating conditions.

Figure 1. ASP Sterrad 100 H_2O_2 Sterilizer

The opening and closing of the sterilizer is software driven via the built-in computer. The samples to be exposed can be laid on the racks within the sterilizer. Alternatively, they can be placed within a standard ASP basket with a cover, which has numerous holes for efficient diffusion of hydrogen peroxide to the interior. Such a tray can be seen within the sterilizer in Figure 1.

The samples are loaded within the basket; however the samples do not directly touch the bottom of the basket. Instead, they are loaded onto a glass tray, which is shown below in Figure 2. In this way, the samples are maximally exposed to the H_2O_2 vapors.

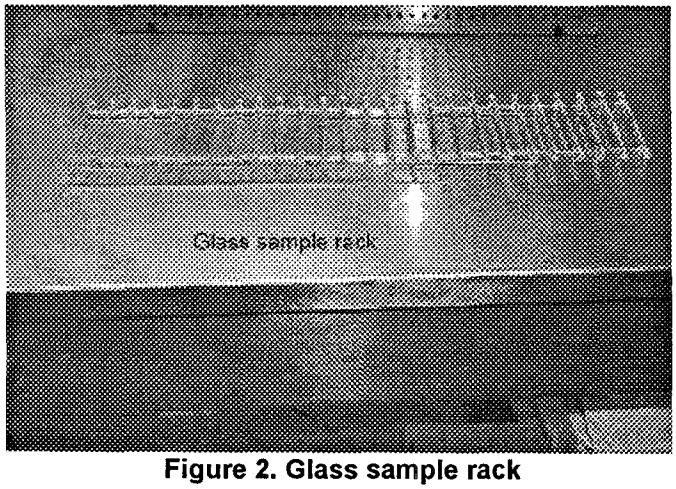

Figure 2. Glass sample rack

Sterilizer Operating Cycle

The sterilizer can be configured to operate multiple cycles for a single experimental run. With each additional cycle, the sample is exposed to more H_2O_2. In a standard hospital sterilization process, 2 cycles are recommended. However, for the purpose of ensuring a thorough exposure of materials being studied, JPL has opted to run 4 cycles for "worst case" exposure to H_2O_2. A standard cycle is explained below with the help of Figure 3.

By referring to Figure 3 it can be seen that there are 5 different phases in a H_2O_2 exposure run.

1) Vacuum – In this stage, the chamber is evacuated to ~ 0.3 mm Hg. This stage takes typically 5-20 minutes. This is the initial stage of all cycles.

2) H_2O_2 injection – In this stage, hydrogen peroxide is injected into the vaporizer bowl by either automatic injection via the automatic cassette mechanism or by manual injection with a syringe via the manual injection port. The chamber pressure will then rise to ~ 5 Torr. A typical value for the H_2O_2 concentration varies between 3-5 mg/liter of chamber volume. Fig.4 below shows the H_2O_2 concentration for a typical cycle.

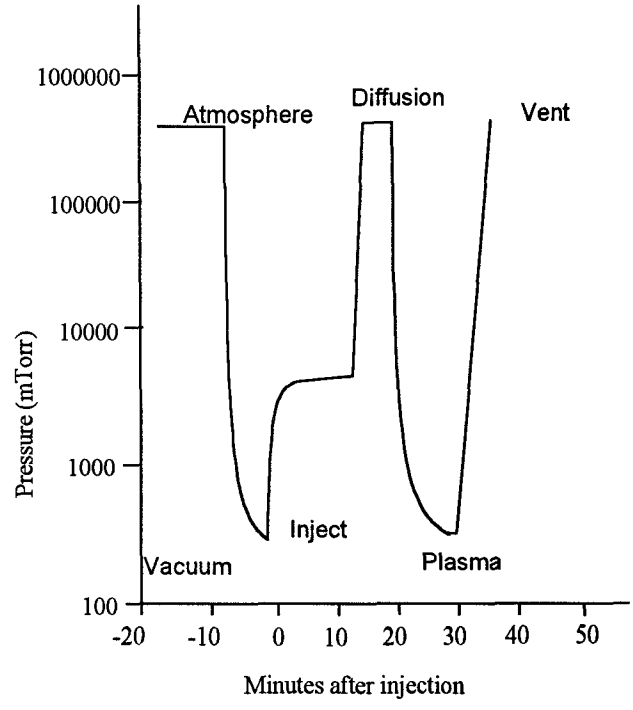

Figure 3. Sterrad 100 SI GMP Sterilization Cycle

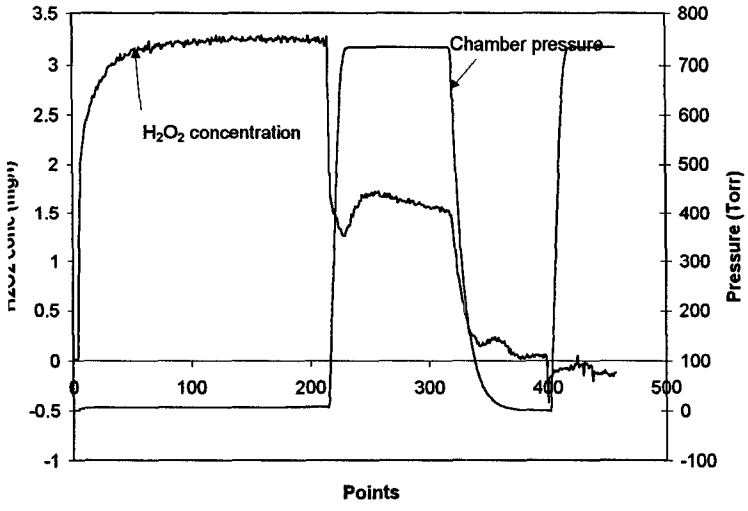

Figure 4. Profiles of hydrogen peroxide concentration and chamber pressure in a typical run.

3) Diffusion stage – this stage allows sterile filtered air into the chamber. By this, the hydrogen peroxide is driven into packaging and diffusion restricted areas and generally enhances the efficacy of the sterilization process.

4) Plasma – low temperature hydrogen peroxide gas plasma is created inside the chamber. Prior to this stage, the chamber is evacuated to ~ 500 mTorr. Plasma is needed to break down the hydrogen peroxide into nontoxic products, namely, water and oxygen. However, plasma is considered harmful for the spacecraft. As a result, the plasma stage is minimized to 1sec.

5) Vent – This stage is the final stage in the sterilization process. Sterilized filtered air is allowed into the chamber.

Some of the parameters that can be adjusted for a cycle include – injection time, diffusion time, plasma time, number of injections, number of plasma cycles, number of diffusion cycles. In addition the 5 stages mentioned above have many parameters that can be tweaked. The current version of software does not allow changing the chamber temperature; however future versions will support changing the chamber temperature.

Operating Envelope – Pressures, Temperatures for Given H_2O_2 Concentration

In order to ensure that the H_2O_2 available for sterilization is always in the vapor phase, it is important that the right operating conditions for pressure and temperature are maintained. If the operating temperature is below the dew point of the H_2O_2-H_2O mixture it is possible to condense out pure H_2O_2, which can be quite deleterious to spacecraft components. A thermodynamic study was conducted to determine the operating envelope. The bounding curves (59 wt.% H_2O_2 at start of run and 0% H_2O_2 at end of run) are shown below in Fig. 5.

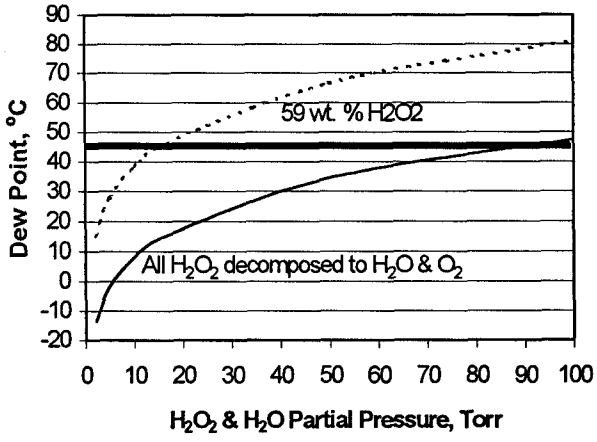

Figure 5. Dewpoint of H_2O_2 and water mixture as a function of chamber pressure

During the injection phase (stage 2), 1.8 mL of a 59 wt.% hydrogen peroxide is injected on to 55°C bowl. The chamber temperature is maintained above 45°C and total chamber pressure at less than 10 Torr (H_2O_2 partial pressure of 4.5 Torr and H2O partial pressure of 5.5 Torr). In our case the sterilization temperature is above the dew point (~38°C) and hence there is no danger of H_2O_2 and water condensation.

During diffusion phase (stage 3), if there is no decomposition of hydrogen peroxide, the partial pressure of H_2O_2 and H_2O will remain the same (10 Torr) and dew point is still 38°C. If hydrogen peroxide is 100% decomposed to H_2O and O_2, the dew point of resulted gas mixture is lowered to ~10°C.

III. STERILIZER PERFORMANCE

Efficacy of Sterilization Process

All of the samples exposed to H_2O_2 vapor were checked to ensure that proper sterilization conditions were met. Both biological and chemical indicators were employed. Chemical Indicator Strips (Sterrad Chemical Indicator Strip REF 14100, Advanced Sterilization Products, Johnson & Johnson Medical Inc.) were used as a quick confirmation that sterilization conditions had been achieved. A red colored indicator strip changes to light yellow or colorless when exposed to H_2O_2 vapor. If inadequate sterilization occurs, the resultant color change is red to dark orange.

Biological Indicators (BI's) are used to further ensure that sterilizing conditions have been met. The BI's are also manufactured by Advanced Sterilization Products and contain 10^6 *Bacillus stearothermophilus* spores

deposited on glass fiber filter paper (7 mm diameter) and packaged in a small Tyvek package. The BI is placed in with the load in the sterilization cycle. After the cycle is complete, BI's are tested for growth in Tryptic Soy Broth at 60°C. Any resulting turbidity of the broth due to growth of the bacteria indicates that sterilization conditions have not been met, and conversely the absence of growth indicates adequate sterilization conditions.

For these studies both the chemical and biological indicators always showed that sterilization conditions were adequate. A single minimal H_2O_2 exposure cycle was occasionally used and sterilization conditions were always met.

H_2O_2 Diffusion

The purpose of this set of experiments was to investigate if hydrogen peroxide can diffuse through a long narrow path under a typical sterilization condition. Figure 6 shows the glass apparatus used to carry out this experiment.

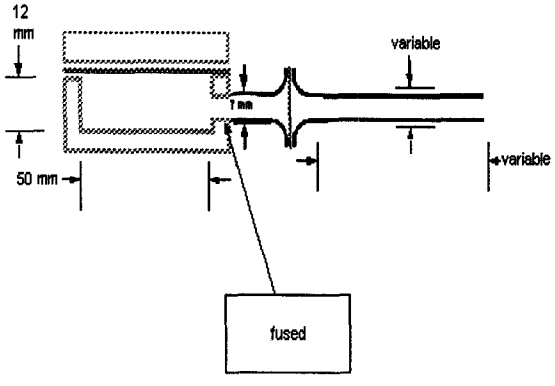

Figure 6. Glass apparatus used for H_2O_2 diffusion experiments.

The apparatus consists of a 30 cm (12 inches) long glass tube connected to a small chamber to hold chemical (CI) and biological indicator (BI) samples. The tube diameters tested were 0.4, 0.8, 3.06, and 5.94 mm. The indicator samples were placed inside the chamber, and it was closed and sealed using an o-ring and a clamp. The entire assembly was placed inside a sterilizer and exposed to one H_2O_2 injection cycle described earlier. In this situation H_2O_2 vapors are forced to the chemical and biological indicators through a 30 cm long and narrow tube. The following four set of experiments were conducted.

1. Indicator control experiment consists of no H_2O_2 exposure. Biological indicators are placed in a nutrient broth to verify positive growth.
2. Growth media control experiment to verify that growth media is sterile.
3. Leak test experiment to verify that chamber does not leak through the o-ring and tube connector.
4. Diffusion experiments with tube diameters varying from 0.4 to 5.94 mm.

The experimental data are shown in Table 1. It is evident that regardless of tube diameter, in all cases sufficient H_2O_2 was able to penetrate to chemical and biological indicators. It is evident that quantity and uniformity of H_2O_2 dispersion is more dependent upon sterilizer operating cycle (H_2O_2 solution injection in vacuum followed by pressurization with air), and methodology used to inject H_2O_2 solution in to a sterilizer chamber than any limitation caused by H_2O_2 diffusion.

Table 1. Hydrogen Peroxide Diffusion Experiment

Diameter, mm	Tube Length, Cm	CI –Result	BI-Results
5.94	30	Met criteria	No growth
3.06	30	Met criteria	No growth
0.8	30	Met criteria	No growth
0.4	30	Met criteria	No growth
Closed end, to check o-ring and tube connector seals	N.A.	Unchanged, red, not sterile	Positive growth
Media control	N.A.	N.A.	No growth
Indicator Control	N.A.	N.A.	Positive growth

Materials Compatibility

A variety of flight hardware materials were exposed to hydrogen peroxide and the changes in material properties were studied [7]. The types of material property tests performed were chosen from end point use criteria. This helped in limiting the number of tests to cover the most important ones. ASTM test procedures were followed in all cases. Samples were tested before and after exposure to sterilization process. The sterilization cycle conditions were the same as discussed earlier. Approximately 50 different types of spacecraft material samples were exposed to four sequential H_2O_2 injection cycles.

Fifteen different sets of metal samples were tested for strength. Where the metal samples were in sheet form, "dog bones" were made for tensile testing. Pre- and post-exposure samples were tested for hardness and under tensile load from which modulus, ultimate strength, yield strength, and strain at yield values were determined. Where the metal samples were in rod form, only ultimate strength values could be determined from the tensile test.

Four thermal control coatings were evaluated for paint adhesion, emissivity and solar absorptance for both pre- and post exposure to H_2O_2. Thermal control coatings are painted onto surfaces to modify and provide specific thermal-optical properties of the surface. They are used as a form of passive thermal control. Good adhesion of the coating is important to provide a surface that is handleable and is not subject to the generation of particulates, which could be sources of contamination for critical components on the spacecraft. These test panels were subjected to thermal cycling to simulate their expected thermal use profile.

Two types of tapes and encapsulants were tested for adhesion. Typical uses of these tapes include closeouts of honeycomb structures to prevent ingress of contaminants, anchoring structures, and securing blankets. One type of cable tie was tested for ultimate load. Velcro was tested for peel strength and particle fallout.

Six thermal blanket materials were tested for emissivity, solar absorptance, coefficient of thermal expansion, and glass transition temperature (T_g). Thermal blankets provide passive thermal control for spacecraft components.

One representative and commonly used type of tin solder was tested for resistivity. Since tin solder provides the electrical connectivity between components, this was the only test performed to see if the resistivity is compromised by H_2O_2 exposure.

Four different composite materials and three types of honeycomb materials were tested. Depending on end-usage, the tests varied. Thus, for G10 and G11, which are used for applications such as non-structural insulation, shims, washers, etc., measurements conducted were modulus, yield strength, ultimate strength and strain at yield. For structural composites such as XN-80/EX1515 and M55J/BTCY1, tests were conducted to determine the modulus, ultimate strength, compressive strength and Iosipescu shear tests. For the 5052, 5056 aluminum, and Hexcel phenolic flex core honeycomb materials, flat-wise tensile tests were performed with FM73 used as the film adhesive. Honeycomb materials are used in lightweight structural components and the flat-wise tensile strength test is the standard reference test for workmanship of the assembled panels. The test determines the face sheet integrity and bonding between face sheets and core.

Only one type of gasket sealing material – Viton 75 was tested for ultimate strength. If the seal hardens or if it takes a compressive set and loses resiliency, a loose-fitting seal might ensue. Since the material is isotropic, one cost-effective test would be to test the tensile strength and from that get a feel for compressive loading as well. Due to cost and time constraints, only this test was performed. Future testing for compressive set and hardness are recommended for a complete understanding of the effect of exposure to H_2O_2.

Three types of plastics were tested – Teflon, Kynar and Ultem. Teflon was tested for modulus, ultimate strength, yield strength and strain at yield. Kynar was tested in the form of a heat shrink tube between two rods. An extensiometer could not be attached and cross-sectional area at point of failure could not be determined. As a result, only ultimate load values could be determined for Kynar. Ultem is a brittle material and it was not possible to obtain yield strength and strain at yield, leaving only modulus and ultimate strength that could be determined.

Only one type of lubricant was tested – Braycote 600. This is an oil which is commonly used in space qualified mechanisms. An ASTM-derived test called the "Inclined Plane Method" for determining the coefficient of static friction was used.

Eight types of polymeric adhesives were tested for lap shear strength. In this test shear loads on the structural bonding are applied and the resulting ultimate shear strength is determined.

The materials compatibility results are summarized in Figure 7. Overall test results are broadly classified under a) no change or change < 15%, or b) significant change when > 15%. When there is a significant change in the material test property, a 25% reduction of pre-exposed value is recommended for design purposes. What this means is that an upper limit for design purposes should not exceed 75% of the pre-exposed value.

Isolation of Sensitive Parts from H_2O_2

Certain electronic parts are very sensitive to any cleaning and/or sterilization process and must be launched contaminated with microorganisms. One way to deal with such parts is to enclose them in a sealed box such that no contamination from within escape, nor any from outside, enter. However, they must be free to vent under the influence of the changes in external pressure experienced in space exploration. This is why HEPA filters are placed over openings in the enclosure, allowing pressure equalization. HEPA filters are fine enough to prevent internal contamination from getting out, but yet allow the passage of gas.

If current H_2O_2 sterilization process is adopted to terminally sterilize spacecraft and lander hardware, it is safe to assume that sterilization chamber is first subjected to evacuation down to 0.3 Torr followed by injection of H_2O_2 solution. The solution immediately flashes to vapor and increases the pressure in the chamber to 5-10 Torr. The vapor is allowed time to penetrate and then filtered air is introduced into the chamber until ambient pressure is reached. This drives H_2O_2 and water vapors into all restricted areas including enclosure interiors where sensitive electronic parts would be located. A well focused task at JPL is

addressing following issues that will lead to redesign of enclosures to accommodate a terminal sterilization process.

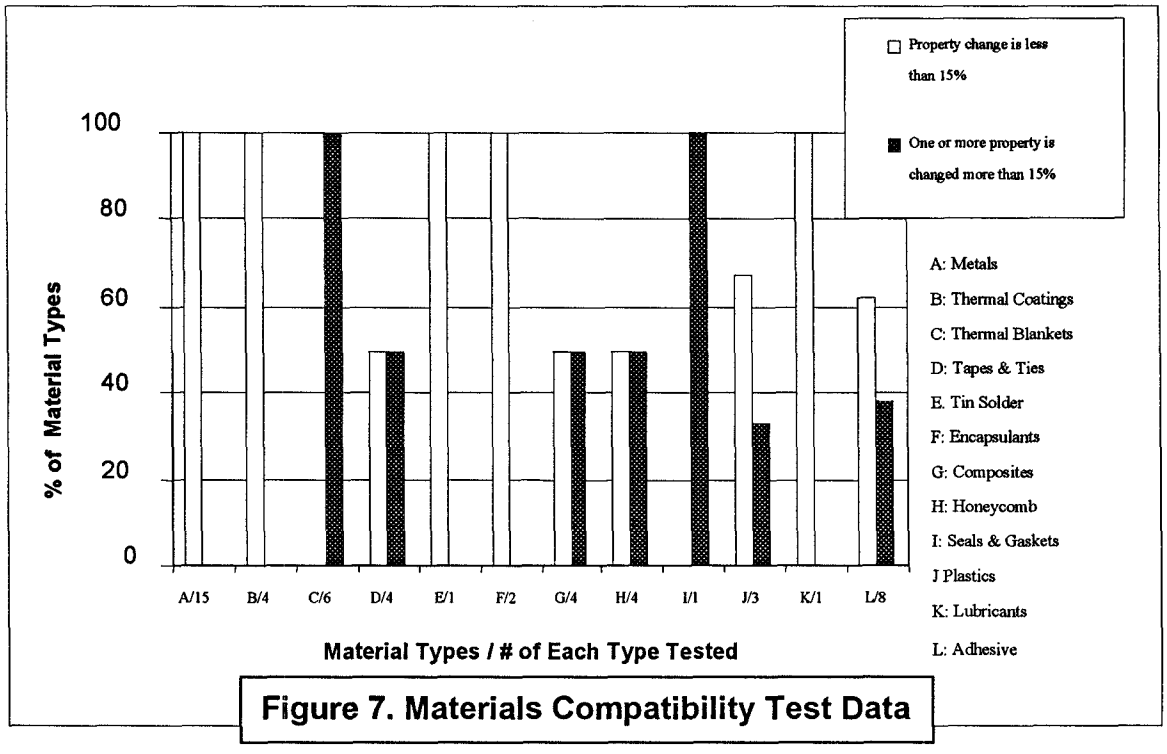

Figure 7. Materials Compatibility Test Data

1. The filter must allow adequate gas passage in either direction such that differential pressure between the inside and outside of the enclosure doesn't exceed the structural capabilities of the enclosure. The introductions of filtered air will be the events that determine the HEPA filter cross sectional area since the differential pressures will be the greatest during these events. The flow velocity during the introduction of H_2O_2 that increases the chamber pressure from 0.3 to 10 Torr is trivial by comparison.
2. The dynamics of H_2O_2 and water vapor flow through HEPA filter under pressure gradients experienced during a sterilization cycle must be well characterized.
3. Based on issue 2 above, we plan to investigate the possibility of first decomposing H_2O_2 to harmless water vapor and oxygen molecules followed by removal of water before oxygen enters into the enclosure interior. This could be through a filter assembly that contains three layers. The layer outermost to the enclosure is one that has a catalyst such as titanium oxide, silver, or platinum on it to break down the H_2O_2 vapor into water and O_2. The catalyst may perhaps be placed on a fine mesh or thin layer of relatively coarse sintered metal substrate. The next layer in is a desiccating material to remove the water vapor leaving only O_2 to continue on. The desiccating material may be a quite porous solid or bag of coarse powder such as silica gel, activated alumina, or activated bauxite. The innermost layer is the HEPA filter itself to prevent bacterial passage into or out of the enclosure. Only gas (air and O_2) moves from inside the enclosure to the outside. But H_2O_2 and water vapor try to move from the outside to inside the enclosure. Consequently, the filter assembly must only decompose H_2O_2 and remove water from the flow trying to enter the enclosure. The catalyst layer may consist of the catalyst wash coated onto a medium. The catalyst can definitely be applied to a sintered metal medium, but it may be possible to apply it to some other lighter medium such as cloth or fiberglass.

This work is under progress at JPL and test results will be discussed in the future publications.

IV CONCLUSION

A vapor phase hydrogen peroxide sterilization process appears to offer a low temperature alternative to the dry heat process to terminally sterilize a spacecraft. The process seems to be effective and compatible with many of the flight hardware materials. A significant amount of research work still has to be done before this process can be certified to terminally sterilize a spacecraft.

V ACKNOWLEDGMENTS

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CONTACT

All inquiries should be addressed to Dr. Naresh Rohatgi, Jet Propulsion Laboratory, Mail Stop 125-224, 4800 Oak Grove Drive, Pasadena, California, 91109 or at naresh.rohatgi@jpl.nasa.gov